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EFFECTS OF POLLUTANTS ON BIOLOGICAL SYSTEMS

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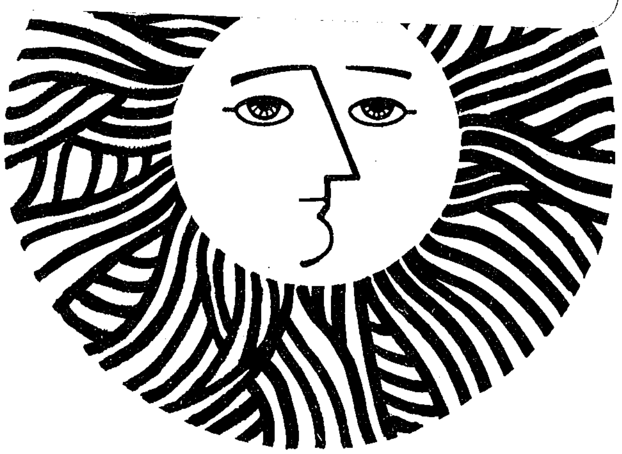

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EFFECTS OF POLLUTANTS ON BIOLOGICAL SYSTEMS

FY-1979

The research reported in this volume was undertaken during FY 1979 within the Energy & Environment Division of the Lawrence Berkeley Laboratory. This volume will comprise a section of the Energy & Environment Division 1979 Annual Report.

CONTENTS

EFFECTS OF POLLUTANTS ON BIOLOGICAL SYSTEMS

INTRODUCTION

D. LEVY

Planning a rational energy future requires anticipating the environmental consequences of various technologies. This is difficult to do with precision as the effects of pollutants are often determined by interactions between and among complex physical (abiotic) and biological (biotic) systems. A given pollutant may affect human beings through direct exposure or indirectly through inducing changes to biological systems which humans need to utilize. The concentration of a toxin in the food chain or the destruction of organisms necessary for the maintenance of high quality water are examples of indirect effects. Pollutants can be transformed and/or degraded as they establish residence in various components of an ecosystem. Anticipation and amelioration of pollutant effects involves the integration of a vast range of data. This data includes:

- physical and chemical characterization of the pollutant as it enters the environment,
- determining effects on the various components (biotic and abiotic) within the context of the functioning ecosystem of interest,
- transformation in movements and/or degradation of the pollutant within that ecosystem and within specific organisms and physical components, and
- determining a detailed biochemical and biological picture of the interactions of pollutants with particular organisms and/or their cellular components judged salient for various processes.

The major programs described below are designed to answer parts of the above fundamental questions relevant to pollutants generated by energy related technologies. Their emphasis is on anticipating consequences to the biological components of various ecosystems. The work ranges from studies involving parts of a single cell (the membranes) to studies involving the whole ecosystem (in the pelagic zone of a lake). The programs take advantage of expertise and technical abilities present at LBL.

Two small exploratory projects which were of brief duration and not related to anticipating biological effects of pollutants are included in this section. They concern geothermal technology and its improvement using techniques based on organic and physical properties of certain materials.

PAREP: POPULATIONS AT RISK TO ENVIRONMENTAL POLLUTION*

D. Merrill, B. Levine, S. Sacks, S. Selvin, and C. Hollowell

INTRODUCTION

The project PAREP (Populations at Risk to Environmental Pollution) supersedes an earlier project PARAP (Populations at Risk to Air Pollution). An integrated data base assembled by the LBL Computer Science and Applied Mathematics Department[1] is being analyzed by the LBL Energy and Environment Division in collaboration with the School of Public Health of the University of California.

The PAREP data base covers the United States and territories at the county level, with subcounty detail for some data elements. The data base consists of socioeconomic and demographic data, mortality data, and air-quality data. Approximately 3,000 data items are available for each county.

Air Quality Data

Previous nationwide analyses have averaged air-quality measurements from the monitoring stations within each county. Such a method is unsatisfactory because (1) many counties have no active monitoring stations, and (2) many people live closer to stations outside their county than to those within their county of residence.

Accordingly, in its task of creating a nationwide directory of air-quality data by county, the PAREP group first determined reliable latitudinal and longitudinal coordinates for each active monitoring station, corrected numerous errors in the existing EPA monitoring station directory file, and combined these data with related files from several independent sources. Discrepancies were resolved by computer, when possible, and by consulting maps.

Yearly summaries of air-quality data for 1974-1976, covering nine pollutants, were obtained from the EPA SAROAD (Storage and Retrieval of Aerometric Data) data bank and merged with corrected coordinates.

Figure 1 displays the three-year geometric mean value of total suspended particulate concentration at monitoring stations in California. As is evident from the figure, air quality is poorest in the Los Angeles area. Air quality was then estimated at the population center of each county as a weighted average of measurements from all nearby stations, whether in the county or not.

Mortality Data

The PAREP data base contains county-level age-adjusted mortality rates[2] by sex and race from two sources: (1) 1968 to 1972 mortality statistics for 53 causes of death, from the University of Missouri; (2) 1950 to 1969 mortality statistics

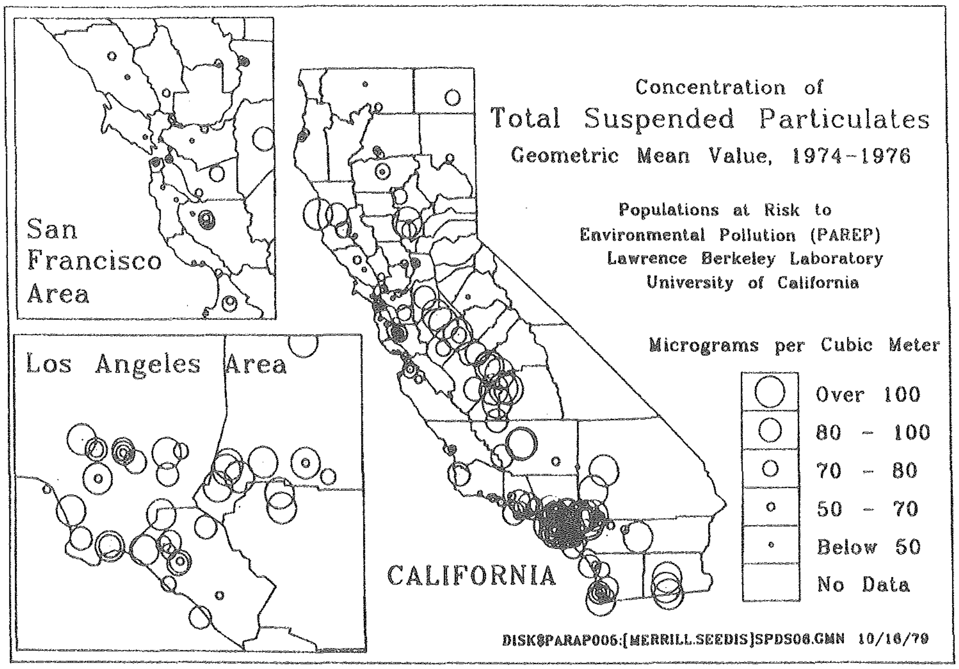

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Fig. 1. Three-year geometric mean value of total suspended particulate concentration is plotted as a circle at the station's location. The size of each circle indicates the relative pollutant concentration.

for 35 cancer sites, from the National Cancer Institute.

Standard scores were calculated as

(county rate - U.S. rate)/(error)

where "error" is the absolute statistical error of the county rate, estimated as the county rate divided by the square root of the number of deaths (or the expected number of deaths, if no deaths occurred).

Table 1 illustrates the incidence of stomach cancer among white males in Arizona for 1968-1972 using this standard score. Although the rate in Maricopa county (around Phoenix) is not as low as in four other counties, its deviation below the U.S. mean is statistically the most significant.

Figure 2 presents similar data for California and Hawaii in map form. Significant deviations above the U.S. mean are observed around Los Angeles, San Francisco, Sacramento, and Hononulu. By plotting standard scores rather than rates, random fluctuations in small counties do not obscure statistically significant trends in large cities.

Socio-economic and Other Data

Indicators of socio-economic status (SES) (income, education, employment by industry and occupation, etc.) were obtained from the 1970 U.S. Census. In addition, the PAREP data base includes corrected 1970 population counts by age, sex, race, and marital status (for the purpose of normalizing mortality or morbidity rates), the Census Bureau's best available 1970 county population estimates, and related files providing subcounty detail on the geographic distribution of the U.S. population.

Most of the PAREP data base is being converted to a format suitable for use in the SYSTEM 2000 Data Base Management System. The same data and associated files are installed in LBL SEEDIS (Socio-economic Environmental Demographic Information System), an interactive information system operating in a network of DEC VAX computers. SEEDIS permits the PAREP data to be used in combination with files from other sources. Selected data, as well as data entered by the user, can be easily combined and displayed in tabular or graphic form, including maps.

Table 1. Stomach cancer in white males, State of Arizona U.S. rate = 10.20.

	1970 Population	1968-72 Annual Rate per 100,000	Standard Score
AN PINAL	29983	15.02	1.43
AN YAVAPAI	17982	15.26	1.16
AZ YUMA	28938	12.24	0.65
AZ APACHE	3913	0.00	-0.00
AZ SANTA CRUZ	6429	10.11	-0.02
AZ GRAHAM	7381	9.68	-0.10
AZ GILA	12066	9.75	-0.12
AS PIMA	160936	10.10	-0.12
AZ GREENLEE	5038	9.00	-0.20
AS MOHAVE	12588	8.36	-0.49
AZ NAVAJO	11683	4.13	-2.18
AZ COCONINO	17359	3.99	-2.77
AZ COCHISE	30222	4.81	-2.89
AZ MARICOPA	448324	7.97	-3.61

ANALYSIS

Analysis of the PAREP data base is in progress in the following areas.

Estimation of Air Quality

This analysis considers the problems involved in geographic interpolation of annual average air-quality measurements. Statistical and graphic techniques are used to determine the validity of the averaging procedures described above. The basic criterion is that the model must accurately predict air quality at the position of a monitoring station as a function of measurements from nearby stations. The study will attempt to determine (1) the best value of the scaling parameter d_0 (described above) and (2) realistic standard deviation errors associated with county air-quality estimates.

Ecologic Patterns of Disease in the United States

The PAREP data base is an ecological data base, i.e., the basic unit is a group of individuals whose characteristics are known only on the average. In other words, analysis of PAREP data will not identify cause-and-effect relationships between air pollution and lung cancer, for example. The data on which the PAREP must rely does not permit such inferences to be drawn. On the other hand, PAREP can provide, at low cost, a quantitative description of the relationships among a large number of variables. Any strong correlations that emerge and cannot be explained would become candidates for intensive study. We are using the straightforward analysis technique of multiple regression, with mortality as the dependent variable and other variables (income, education, air quality, etc.) as independent variables. Patterns of disease are analyzed after removal or partial removal of the hypothesized linear influences of SES and air-quality variables. Such an analysis was performed earlier[3,4] on a preliminary data base containing California data. Statistical limitations prevented any firm conclusions from being drawn. A similar analysis is being repeated for the entire United States.

As part of the PAREP project, we are also analyzing data from the Third National Cancer Survey, which recorded all incidences of cancer between 1969 and 1971 in nine areas of the United States. Individual case records, coded by census tract, have been merged with tract-level SES data from the 1970 census, and tract-level air quality estimates calculated as described above. Multiple regression and other techniques are being used to describe relationships among air quality, socio-economic status, and the incidence of certain histologic cancer types.

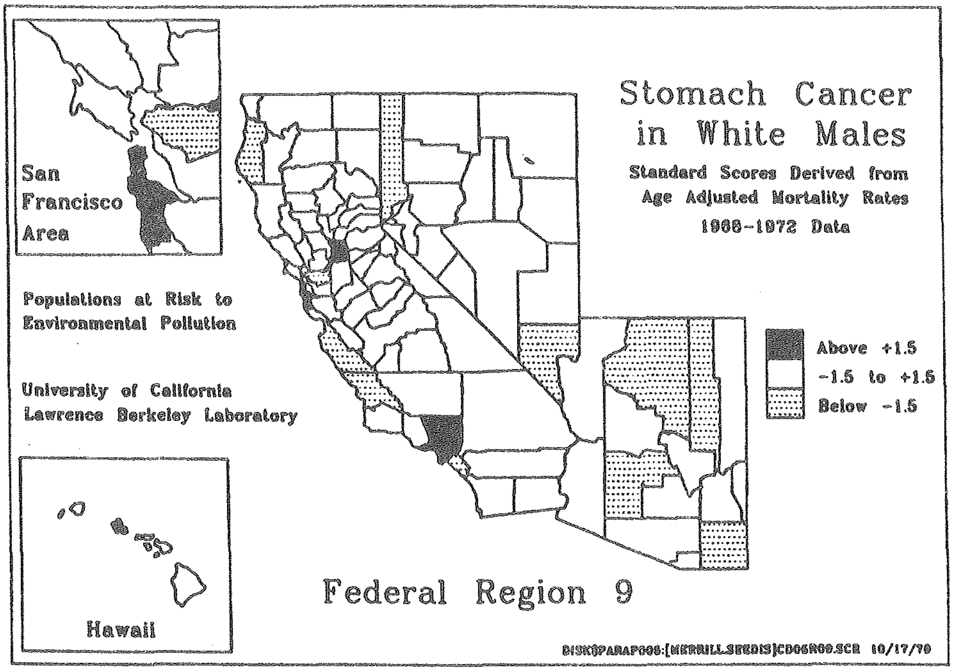

Fig. 2. Typical of maps used to display geographic correlations in mortality statistics.

PLANNED ACTIVITIES FOR 1980

The following projects, to be described in terms of data, general analytic strategy, and potential future direction, are proposed for 1980.

1. Ecologic patterns of disease in the United States.
2. Air quality, socio-economic status and cancer incidence in the San Francisco Bay Area.
3. Consequences of ecologic regression.
4. The association of socio-demographic measurements and histologic cancer type for nine sites.
5. Standardization of age-adjusted mortality rates for county-level data.

FOOTNOTE AND REFERENCES

*The work described in this report was funded by the Office of Health and Environmental Research, Assistant Secretary for Environment of the U.S. Department of Energy under Contract No. W-7405-ENG-48.

1. S. T. Sacks, S. Selvin, and D. W. Merrill, Building a United States County Data Base: Populations at Risk to Environmental Pollution, presented at the National Institute of Aging, Bethesda, Md., June 26, 1979; Lawrence Berkeley Laboratory Report, LBL-9636 (September 1979).
2. S. Selvin, S. T. Sacks, and D. W. Merrill, "Standardization of age-adjusted mortality rates," Lawrence Berkeley Laboratory Report, LBL-10323 (February 1980).
3. D. W. Merrill, S. T. Sacks, S. Selvin, C. D. Hollowell, and W. Winkelstein, Jr., "Populations at Risk to Air Pollution (PAREP). Data base description and prototype analysis," Lawrence Berkeley Laboratory Report, UCID-8039 (August 1978).
4. C. D. Hollowell, S. T. Sacks, S. Selvin, B. S. Levine, D. W. Merrill, and W. Winkelstein, Jr., "PAREP: Populations at Risk to Environmental Pollution- Data base description and prototype analysis," in Energy and Environment Division Annual Report 1978, pp. 152-155; Lawrence Berkeley Laboratory Report, LBL-8619 and UC-13 (1979).

MEMBRANE OXIDATIVE DAMAGE*

L. Packer, A. Quintanilha, E. Kellogg III, R. Mehlhorn,
M. Nova, L. Cheng, and J. Maguire

INTRODUCTION

We are pursuing studies on the effects of environmental and polluting factors on mammalian cellular membranes. Our previous reports[1,2] showed how sensitive isolated mitochondria, whole cells (Hepatocytes), and model membranes are to the damaging species of oxygen.

We have assumed that ozone and various reactive species of oxygen, some of which could be generated by several cellular photosensitizers in the presence of visible light, will react with enzymes and other protein and liquid constituents of membranes thereby altering their activity and structure.[3] Our studies have been extended to <u>in vivo</u> and <u>in vitro</u> studies of light sensitivity of catalase, a crucial enzyme involved in the protection against oxidative damage. Since catalase is a key enzyme in H_2O_2 metabolism, the importance of its inactivation to the overall metabolic protective capacity of cells is at present being carefully characterized to identify its significance in the time sequence of cellular damaging events. Superoxide involvement in the elusive bactericidal effects of negative air ions has also been investigated. Spin trapping techniques have been used to assay for different types of radicals, and model systems have been perfected to monitor membrane protein and lipid-protein interactions to allow us to localize and characterize the mechanisms of oxidative damage.

CATALASE STUDIES

Catalase, an enzyme consisting of four heme containing subunits, decomposes H_2O_2 and may be one of the main defenses against oxidative toxicity in the cell. Figure 1 shows the main physiological pathway in which catalase is involved. We have described the inactivation of catalase <u>in vivo</u>, in intact isolated hepatocytes exposed to visible light.[4] Photoinactivation of catalase with visible light (>400 mm) was also done in peroxisomal catalase in the mitochondrial fraction of rat liver, and in purified bovine liver catalase.[5]

The action spectrum (Fig. 2) indicates that light with a wavelength corresponding to that of maximal absorbance of the heme moeity acted most effectively in the photoinactivation of catalase. The O_2 dependence of catalase photoinactivation shows that high concentrations of O_2 stimulate inhibition, while anaerobic samples are not inactivated. Scavengers of $\cdot OH$ (0.25 M sucrose), $^1O_2$ (1 mM histidine) and O_2^- (10 μg/ml SOD, superoxide dismutase) did not prevent the inactivation process, while catalase substrates (100 μM formic acid, methanol or ethanol) afforded complete protection.

Peroxisomal catalase in the mitochondrial fraction is far more susceptible to photoinactivation than purified catalase; much lower light intensities are required for inactivation in the mitochondrial system, but superoxide dismutase does afford partial protection. Our results indicate that despite the strict dependence upon O_2, no protective effect of scavengers of $\cdot OH$ and $^1O_2$ could be detected. This lack of protective effect may result from the inaccessibility of these scavengers to the active site. The complete protective effect of substrates may involve their scavenging of $\cdot OH$ at the active site itself. The fact that in the case of the substrate formic acid, only 0.1% of the hemes are ligated at any time, suggests that the protective effect of substrates

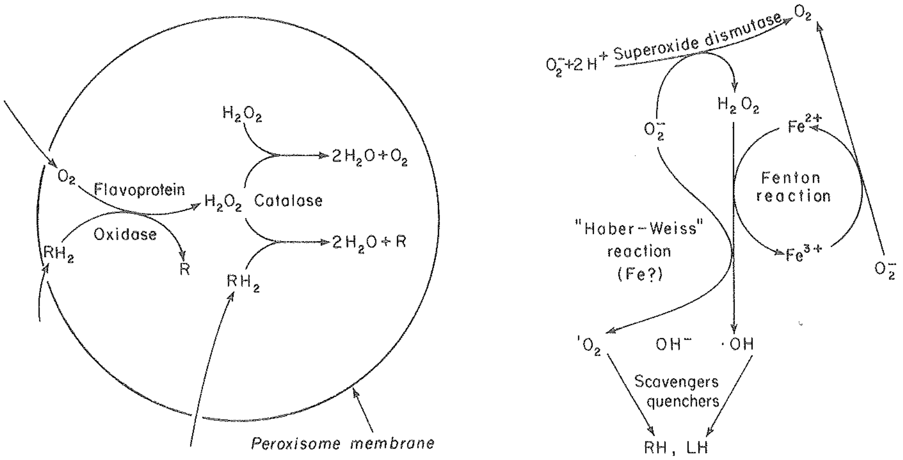

Fig. 1. Chemical and biochemical reactions for the formation and removal of peroxides. (XBL 809-1920)

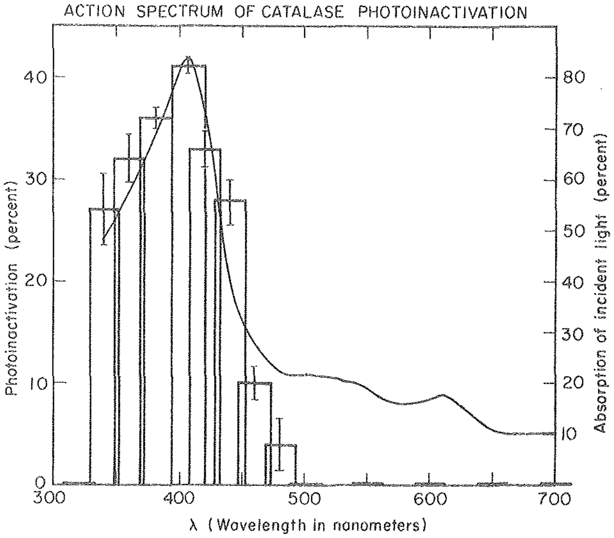

Fig. 2. Action spectrum of catalase photoinactivation. (XBL 793-3330)

results from their conversion of catalase from Compound I to the Ferricatalase form[6] and a marked photosensitivity of Compound I alone.

The protection of catalase of the mitochondrial fractions by superoxide dismutase suggests that O_2^- and H_2O_2 may be produced by the mitochondria or the peroxisomes during illumination. The physiological importance of the photoinactivation of catalase may be particularly important in "Acatalasemic" individuals exposed to short term light stress. Catalase inactivation would result in large increases in H_2O_2, thereby promoting the formation of ·OH, a dangerous oxygen radical. Recent work[7,8] has shown that the wavelength responsible for chromosomal damage peaked at 405 nm, the wavelength that corresponds to the maximal absorbance and inactivation of catalase in our studies. These results, and the fact that externally added catalase eliminated the light induced chromosomal damage, suggest that the light damage seen in their systems may result mainly from catalase inactivation.

SUPEROXIDE EFFECTS ON BACTERIA

The underlying biochemical changes of the biological effects of small air ions have only been detected in a few instances.[9] Many studies have demonstrated the bacterial effects of O_2^-. Since the mysterious physiological and psychological effects of negative air ions have attracted the interest of many investigators, we decided to test for the involvement of O_2^- in this phenomenon by evaluating the protective effect of SOD.[10]

We found that exposure of bacteria to negative air ions caused total loss in viability. The addition of 10 µg/ml SOD protected bacteria completely while catalase or denatured SOD had no significant protective effect. A possible mechanism of the observed effect might involve O_2^- acting as a nucleophile on the phospholipid bilayer, causing a de-esterification of fatty acids. This could lead to an increase in surface charge and a weakening of the membrane, which under hypotonic conditions might lead to cell lysis and death. These results support the concept that the negative air ions responsible for bacterial death must either be hydrated superoxide anions or must generate O_2^- in the suspension medium.

MODEL STUDIES

Spin trapping techniques have been used to study the effects of both biological chelators (such as transferrin and ferritin) and synthetic chelators (such as ethylenediamine tetra acetate, EDTA and diethylenetriamine penta acetate, DETPA) on the Fenton reaction (Fig. 1), where iron reacts with hydrogen peroxide to produce hydroxyl radicals (probably the most potent biological toxicant found *in vivo*). Our results show that biologically bound iron in ferritin has no significant pro-oxidative effect, while iron bound to EDTA has a somewhat enhanced pro-oxidative effect compared to free iron. Other studies involving iron catalysis of epinephrine (a neuro transmitter) autooxidation show a similar pattern of enhancement or suppression of pro-oxidant catalysis by various forms of bound iron. Since iron has been shown to be a powerful oxidant and α-Tocopherol (Vitamin E) is a well known antioxidant, a series of studies is in progress on the protective or damaging effects of different levels of Fe and Vitamin E in controlled diets fed to rats. We have found that iron deficiency decreases the total amount of Fe-S centers in mitochondria and that Vitamin E deficiency increases lipid peroxidation in isolated cells (hepatocytes) and cellular organelles. The effect of Vitamin E deficiency on mitochondrial membranes seems to indicate an increase in their overall negative surface charge which may be related to the presence of greater levels of lipid peroxides found in these membranes. Sucrose (a known ·OH radical scavenger) in the incubation medium, decreases the effects of Vitamin E deficiency on the photooxidative damage to lipids and enzymes of mitochondria. The photo-destruction of spin labels has been used successfully to monitor the release of flavins from biological membranes. Our results indicate that a possible mechanism for mitochondrial photoxidative damage may be the release of important flavin co-factors in some of the dehydrogenase complexes found in these membranes. Several techniques (spin labeling and cross-linking) have been perfected in our laboratory for studying the importance of peptide mobility on enzymatic activity and electron transport in mitochondria and for determining the orientation of intrinsic membrane components.[11-15]

PLANNED ACTIVITIES FOR 1980

Future studies will concentrate on four major areas:

1) To arrive at an understanding of photosensitive mechanisms of oxidative damage in cells and to determine the specific oxygen species responsible for such damage. Other metabolic pathways that involve hemes and flavins (both believed

to be important photosensitizers) such as the drug-metabolizing mixed-function cytochrome P-450 oxidase and other amino oxidases will be studied.

2) To use spin-trapping and spin-destruction techniques to identify the radicals and radical generating species in *in vitro* membrane systems that are responsible for the damage observed.

3) To continue the use of controlled diet studies in rats to modify the composition of the biological membranes under study and to help us in determining the components that enhance or reduce photoxidative damage. Dietary iron and Vitamin E levels will be controlled in those studies.

4) To continue spin-labeling and other EPR techniques which play a major role in the study of such parameters as membrane permeability and fluidity and which will be used to measure membrane surface charge, peptide mobility and the concentration of free radical intermediates in many of the membrane associated enzymatic reactions.

FOOTNOTE AND REFERENCES

*This work was supported by the Department of Energy, Secretary of Environment.

1. *Energy and Environment Division Annual Report-1977*, Lawrence Berkeley Laboratory report, LBL-6877, pp. 142-146 (1977).
2. *Energy and Environment Division Annual Report-1978*, Lawrence Berkeley Laboratory report, LBL-8619, pp. 166-170 (1978).
3. L. Packer and E. W. Kellog III, "Photooxidative damage to mammalian cells and proteins by visible light" in ACS Symposium Series *123*, *Chemical Deterioration of Proteins* (J. R. Whitaker and M. Fujimaki eds.) pp. 83-93, 1980, Lawrence Berkeley Laboratory report, LBL-9672 (1979).
4. L. Y. L. Cheng, and L. Packer. "Photodamage to hepatocytes by visible light," FEBS Lett. *97*, 124-178 (1979).
5. L. Y. L. Cheng, E. W. Kellogg III and L. Packer. "Photoinactivation of catalase by visible light," submitted to Photochem. Photobiol., Lawrence Berkeley Laboratory report, LBL-9743 (1979).
6. G. R. Schonbaum and B. Chance, *The Enzymes*, Vol. XIII, pp. 363-408 (1976).
7. R. Parshad, K. K. Sanford, G. M. Jones, and R. E. Tarone, Proc. Natl. Acad. Sci. USA, *75*, 1830-1833 (1978).
8. R. Parshad, K. K. Sanford, G. Taylor, R. E. Tarone, G. M. Jones, and A. E. Baeck. Photochem. Photobiol. *29*, 971-975 (1979).
9. A. P. Krueger, and E. J. Reed. Science *193*, 1209-1213 (1976).
10. E. W. Kellogg III, M. G. Yost, N. Barthakur and A. P. Krueger. "Superoxide involvement in the bactericidal effects of negative air ions on *staphylococcus albus*," Nature *281*, 400-401 (1979).
11. M. Swanson, and L. Packer. "Effect of cross-linking on mitochondrial cyctochrome *c* oxidase," submitted to Arch. Biochem. Biophys. (1979).
12. R. Mehlhorn, and L. Packer. "Membrane surface potential measurements with amphiphilic spin labels," *Methods in Enzymology*, Vol. LVI, Part G, pp. 515-526 (1979).
13. A. T. Quintanilha, M. S. Swanson and D. D. Thomas. "Rotational mobility of soluble and reconstituted cytochrome oxidase," Biophys. J. *25*, 170a (1979).
14. M. Swanson, R. Mehlhorn, L. Packer, and P. Smith, "An imidoester spin-label probe of cytochrome *c*-cytochrome oxidase interactions," submitted to Arch. of Biochem. Biphys. (1979).
15. M. Swanson, A. T. Quintanilha, and D. D. Thomas. "Protein rotational mobility and lipid fluidity of purified and reconstituted cyctochrome *c* oxidase," J. Biol. Chem. *255*, 7494-7502 (1980), Lawrence Berkeley Laboratory report LBL-9661 (1979).

LAKE ECOTOXICOLOGY: METHODS AND APPLICATIONS*

J. Harte and D. Levy

INTRODUCTION

The complexity of ecological processes and the wide range of potentially toxic substances that can be released into the environment from energy-related activities presents the ecotoxicologist with an enormous task. The purpose of this research is to develop and apply improved methods for predicting ecological effects of toxic substances in freshwater lakes. Part of our research is aimed at understanding the behavior of laboratory microcosms and improving their design and operation to make them more useful, while a second part focuses on applications of laboratory microcosms to selected problems in ecotoxicology.

In an ecological context, a laboratory microcosm is simply a collection of chemicals and organisms within well-defined spatial boundaries, usually under controlled physical conditions, in a volume convenient for laboratory study. The ease with which aquatic microcosms can be perturbed or manipulated, and the opportunity they provide for replication and control of systems with considerably more diversity than single organism cultures, suggests that they may be a useful tool for work in ecotoxicology. However, a number of drawbacks are implicit in their small size and artificial environment, and these drawbacks may drastically limit the range of assessment studies that can be performed reliably. Results obtained from microcosm studies may in some, or perhaps most, cases bear little relation to what would be observed in the natural systems that the microcosms were designed to describe. Even when microcosms are initiated from nearly identical conditions, their replication may not be faithful. Moreover, surface effects may complicate the analysis of nutrient budgets and pollution pathways. Over the past four years we have undertaken a series of studies on microcosm design and operation in order to understand these limitations, and at the same time to improve the effectiveness of microcosms as assessment tools in ecotoxicology.[1-3] These studies utilized microcosms ranging in size from 2 to 700 liters and containing a diverse assemblage of biota initially obtained from local lakes.

One of the most serious problems in using microcosms is the excessive growth of algae on the inner walls of the containers. Because the surface-to-volume ratio of a microcosm is considerably larger than that of a natural lake, surface growth of algae and bacteria can exert a disproportionate, or unnatural, influence on water-column nutrient concentrations and pelagic communities. In addition to the distorting influence of this effect, reliable sampling of the sides and bottom is very difficult to perform. Thus, if the most frequent and reliable measurements are in the water column, then much of the activity in the microcosm is missed during periods of heavy side growth and efforts to balance nutrient budgets will be futile. In FY 78, we demonstrated that surface growth could be eliminated successfully from our systems by the simple expedient of transferring the contents of each system to a clean vessel at weekly intervals. This process of periodic transfer introduces periodic mixing in the microcosms, which could affect biological or chemical phenomena and, potentially, result in a loss of replicability. In an ongoing series of experiments, possible side-effects of this surface-growth-control strategy are being investigated. We have also concentrated on determining how well lake microcosms behave like, or "track", the natural lakes that supply the microcosm water. The goal of this phase of our research is to learn how to design and operate the microcosms so as to increase the resemblance between them and their "parent" lake, with the proviso that the systems developed are not prohibitively complex or expensive. To this end, we have closely integrated our laboratory studies with field investigations of the parent lakes.

As our understanding of microcosm behavior and our ability to enhance microcosm design and operation have progressed, we have initiated experiments relevant to current ecotoxicological problems. One such problem is the alteration of decomposition processes in aquatic ecosystems; such alteration can have a great effect on nutrient cycles and therefore on primary productivity. An ongoing application of our microcosms is the investigation of effects of pollutants on the ability of microorganisms to decompose and mineralize organic detritus. A new research program was organized this year to investigate the impact of acid precipitation on aquatic ecosystems in the Sierra Nevada. It includes two phases: 1) background water quality measurements of selected watersheds on the western slope of the Sierra Nevada, and 2) microcosm research simulating ecosystem response to additions of acid precipitation. Emphasis is placed on acid-metal interactions as the possible mechanism for toxic effects on biota.

ACCOMPLISHMENTS DURING 1979

Two experiments comparing in detail a particular lake's behavior over several months with the microcosms derived from it were carried out.[4] Each experiment utilized a different lake. Field sampling was accomplished using a 14 foot boat equipped with a non-polluting electric motor and standard field sampling equipment. The variables mesured in both lakes and microcosm systems on a weekly basis were pH, illumination levels, fluorescence, inorganic nutrients, and phytoplankton and zooplankton species (volume and numbers). Our microcosms are housed in a temperature-controlled room, illuminated by banks of high-output fluorescent lights on a 12h:12h light:dark cycle. The light irradiance at the surface of the water is now set at 7.0 watts/cm^2 (PAR). Cylindrical containers are used for the microcosms, with the larger containers made of nalgene or fiberglass and the smaller systems made of glass.

In the first experiment, a small reservoir (Lafayette, CA) was compared with replicates of 4 liter and 200 liter microcosms. The reservoir is well used for boating and fishing, and receives substantial run-off after rains. The tracking of this reservoir by the microcosms was very poor. The dominant phytoplankton in the lake throughout the run were diatoms whereas in the microcosms the diatoms disappeared within the first two weeks. Subsequently, flagellates and filamentous greens become the dominant phytoplankton in the 200 liter microcosms. The 4 liter microcosms did not support any significant phytoplankton populations after the first two weeks. Total zooplankton volumes in the microcosms were higher than in the lake. During and subsequent to rain fall, high levels of NH_4^+ (probably due to run off) were found in the lake as compared to the microcosms.

In the second experiment, a considerably larger reservoir (Briones) was compared with replicate -- 15, 50, and 150 liter microcosms. This reservoir is a deep (200 ft) lake which receives little run-off. Its water comes, via aqueduct, from the Mokelumne River of California. One set of the 50-liter microcosms received a weekly 1-liter inoculation of water from the reservoir (gathered on sampling days) while the other 50 liter set did not. During the first 6-7 weeks, tracking of the lake by all the microcosms was excellent, with diatoms, Phacus, and other flagellates the dominant phytoplankton. In addition zooplankton and inorganic nutrients in the microcosms tracked the lake well. After 7 weeks, diatoms in both the lake and microcosms disappeared. After this period, the dominant phytoplankton in the lake were dinoflagellates and blue-green algae, while in the 50 and 150 liter microcosms an attached filamentous green (Ulotrix) dominated. In the small 15 liter systems, a green algae (Gloeocystis) dominated after 7 weeks. Similar behavior was observed within replicate microcosms.

From the results of these experiments and other tracking studies currently in progress, we will extract implications for microcosm users. Preliminary analysis of the data from the completed experiments suggests that tracking may be inadequate in lakes that are not sufficiently large to reduce the relative effects of the surrounding watershed on their behavior, and it may also be inadequate in microcosms that are too small. Tracking difficulties also can arise when diatom populations are important in the natural lake, for these populations do not do well under microcosm conditions. Under suitable conditions, excellent tracking is attainable over periods of 6-7 weeks. The importance of careful taxonomic data collection, as well as water chemistry measurements, is underscored by these studies. If care is taken in the initiation process, replication of results among systems set up under nearly identical conditions is attainable. All of these tentative conclusions are subject to the proviso that they apply to the planktonic, or water-column, studies undertaken to date. Their extension to microcosms with benthic communities remains to be shown, and that is a major aspect of our planned work in FY 1980.

In the acid-precipitation research project[5] progress was made in both field studies and laboratory microcosm work. Field investigation included collection and characterization of 14 western slope lake samples in terms of pH and ambient levels of total and dissolved metals in the water column. These measurements allow for the identification of those lake basins particularly sensitive to acid inputs. such data will be made available for consideration in energy facility siting decisions in California.

Using both 4-liter and 50-liter microcosms, simulated Bay Area (Briones) and Sierra (Dark Lake) lake systems, with and without sediment, have been established and then disturbed by the addition of synthetic acid rain. Changes in zooplankton and phytoplankton species and numbers have been tracked following these perturbations. Changes in metals levels (Al, Cd, Cu, Fe, Mn, Pb, Zn) in the water column of these microcosms have been measured using graphite-furnace atomic absorption spectrometry techniques. Fluctuations in levels of Al, Cu, Pb, and Fe are particularly significant in the systems with sediment, with 10-100 fold increases in dissolved metals levels being observed following acid additions.

Progress has been made in designing and applying small microcosms useful for studying detailed patterns of succession in simple planktonic systems in which the species and numbers of plankton, and the nutrient concentrations, are all adjusted at the outset.[6] These systems are designed to have precisely similar light levels, and are constructed so as to minimize inhomogeneities and other complications arising from mixing patterns and surface growth.

A description of earlier work on the analysis of a practical measure of ecological stability was published in a theoretical paper.[7] It contained proof of a rigorous relation between the value of this measure and the distribution of eigenvalues of the community matrix. Other theoretical studies in progress include an extension of the fluctuation-dissipation theorem from classical mechanics to an ecological context, and development and analysis of models of the interactions of microbes with detritus leading to an understanding of the results of experiments on decomposition that we have carried out.

The fluctuation-dissipation theorem relates the correlations in time of the fluctuations of a stochastic system to the recovery rate from a disturbance of such a system. One of us has shown that currently favored ecological models of population fluctuations in a stochastic environment predict behavior in accordance with the fluctuation-dissipation theorem, despite the fact that the assumptions needed for a rigorous justification of the theorem are not satisfied by these models.[8] Phenomena observed by us in a series of decomposition experiments,[9] which studied the response of lake water samples to detritus additions, are in conflict with models traditionally used to describe the kinetics of microbe-detritus interactions. Using mathematical models, we have shown that the phenomena can be accounted for by assuming the existence of a density-dependent, carrying capacity limitation on the ability of microbes to immobilize mineralized nutrients for their growth.[10] The importance of this coupled experimental and theoretical development is that it may lead to a relatively simple, reliable method for measuring density-dependence effects in microbial populations. These effects are believed to provide an indicator of robustness of ecological nutrient cycles.

Finally, several reviews and assessments of the ecological impacts of energy activities, with an emphasis on water-related damages, were prepared. A paper[16] sponsored by the University of California's Water Resources Center on water resource constraints on energy development in California was published in 1979. Another paper,[12] presented at an international conference on resources at IIASA, Vienna, described a framework for analysis of ecological risks of water-intensive energy technologies.

PLANNED ACTIVITIES FOR 1980

In order to improve the realism of microcosms and their effectiveness as tools for ecotoxicology, study of two microcosm design features will be initiated in FY 80: (1) the establishment of proper mixing rates in the microcosms, and (2) the inclusion of benthic sediments.

Effects of mixing have been studied in detail for estuarine microcosms by other groups,[13,14] who find that microcosm behavior can be quite sensitive to the degree of water-mixing, with unmixed systems often bearing little resemblance to the natural parent system. In a series of tracking experiments involving Briones Reservoir, we will investigate the degree to which tracking depends upon the rate of water mixing in our microcosms. Mixing rates in the lake and the microcosms will be compared using gypsum dissolution measurements. A simple motor-driven paddle wheel assembly will be used to create mixing in the microcosms.

The benthic zone plays an important role with respect to nutrient recycling in most fresh-water lakes. It can also function as a temporary sink for many contaminants which initially find their way into the water column. In shallow non-stratified lakes, such exchanges occur more rapidly than in stratified deeper lakes where turnover times on the order of a year are common. Inclusion of a benthic compartment in a shallow lake-like microcosm is difficult. If the benthic sediments cover the entire bottom, the ratio of benthic surface area to overlying water volume (BS/WV) is often at least an order of magnitude greater than that of most natural systems and so the benthic compartment in this case would exert much too large an effect on the overlying water. Even if the benthic compartment's surface area is reduced so that the ratio (BS/WV) is realistic when compared to a particular lake, mimicking realistic aeration, mixing and dilution may be difficult, particularly when examining models of stratified lakes. Benthic compartments scaled approximately to the size of microcosm containers will be filled with benthic materials from the study lakes. Two variables characterizing the benthic compartment will be studied--the benthic surface area and the rate of water flow across the benthic surface, taking advantage of design features similar to those used by Perez et al.[13] The degree to which water-column chemical concentrations and population levels track those in the parent lake will be monitored.

In order to investigate effects of toxic substances on lake decomposition rates using a method developed by our group,[9] large quantities of highly concentrated dissolved and particulate lake-water detritus are needed. We will develop suitable methods for preparing and concentrating detrital materials of various characteristic size spectra and chemical constituency.[15]

Changes in decomposition activity induced by toxic substances can alter the spectrum of molecular sizes in the detritus pool and cause the fractions of the pool with various degrees of lability to change. To investigate the extent to which altered decomposition activity can affect the constituents of the detritus pool, we will explore the possibility of obtaining a characteristic "fingerprint" of detritus, adapting a method developed in the atmospheric aerosol research group at the Lawrence Berkeley Laboratory for studying the components of materials trapped on air filters. Our approach will consist of measuring separately CO_2 and NO_x evolution from detritus samples that are subjected to increasing temperatures, ranging from ambient to about 800°C. The fingerprint consists of the curve of gas evolution as a function of temperature. A wide range of water samples from California lakes will be fingerprinted. This technique will also be used in the benthic sediment studies described above to characterize changes in the organic constituents of benthic sediments that have remained in laboratory microcosms for various lengths of time. Comparisons with sediment fingerprints from the parent lake will be made.

Both field and laboratory studies on the ecological impact of acid precipitation will be continued. Further characterization of lakes located on the western slope of the Sierra and downwind of major population centers will be pursued through the analysis of sediment and watershed soil samples for metals content. These measurements will aid in the selection of those lakes considered to be most susceptible to damage due to metals resuspension. Additional lake water samples will be collected in the Spring, with emphasis on collection during the period of peak snowmelt when an acid "pulse" may be detected.

Laboratory microcosms will be set up at the Aquatic Microcosm Facility at LBL using water and sediment collected from additional Sierra lakes and will be subjected to successive lowering of pH. Metals levels in the water column and changes in zooplankton and phytoplankton species and numbers will be tracked to determine the biological consequences of acid inputs to simulated aquatic systems.

Theoretical investigations of models for microbial-detritus interactions will continue, with the main emphasis on developing methods of determining basal decomposition rates from measurements of enhanced decomposition activity as stimulated by additions of detritus. Further exploration of the fluctuation-dissipation theorem in an ecological context will also be pursued, with stress on trying to understand the extent of, and the reasons for, its apparent applicability to ecosystem models. An effort will be made to design a suitable experimental test of the theorem in laboratory microcosms.

In order to extrapolate results on ecological impacts of toxic substances from laboratory microcosms to a field situation, it is necessary to consider the fact that lake microcosms are inherently different from natural lakes. The types of variables that the public are generally most concerned with in real lakes, such as the health of sport fish populations, odor, and water clarity (which we call macrovariables), do not lend themselves to direct investigation in the laboratory. Nevertheless, the types of ecosystem components most amenable to laboratory study, such as phytoplankton, zooplankton, and water chemistry (which we call microvariables) often exert a great influence on the macrovariables. In a review article prepared for a National Academy of Sciences study, we will describe the limitations and opportunities in current ability to extract results about macrovariables from information obtained about microvariables from microcosms.[16]

FOOTNOTE AND REFERENCES

*This work was supported by Electric Power Research Institute and the California Policy Seminar.

1. A. Jassby, M. Dudzik, J. Rees, E. Lapan, D. Levy, and J. Harte, "Production cycles in aquatic microcosms," Environmental Protection Agency, Office of Research and Development Research Report EPA-600/7-77-097, National Technical Information Service, Springfield, VA, 51 (1977).
2. M. Dudzik, J. Harte, A. Jassby, E. Lapan, D. Levy, and J. Rees, "Some considerations in the design of aquatic microcosms for plankton studies," Intern. J. Environmental Studies, 13, 125-130 (1979).
3. J. Harte, D. Levy, J. Rees, and E. Saegebarth, "Making microcosms an effective assessment tool," Proceedings from the symposium on microcosms in ecological research, John Giesy ed., (to be published) (1979).
4. This work was carried out by J. Harte, D. Levy, J. Rees, and E. Saegebarth.
5. This work was carried out primarily by K. Tonnessen, a graduate student in the Energy and Resources Program, with assistance by J. Oldfather and G. Lockett.
6. M. Dudzik, a graduate student in biophysics, has been carrying out this effort.
7. J. Harte, "Ecosystem stability and the distribution of community matrix eigenvalues," in Theoretical Systems Ecology, E. Halfon, ed., pp. 453-465, Academic Press (1979).
8. This work was carried out by J. Harte.
9. J. Harte and D. Levy, "Investigation of the responses of lake waters to detritus additions," Lawrence Berkeley Laboratory report, LBL-9314, to be submitted for publication (1979).
10. This work was carried out by J. Harte, D. Levy, and E. Saegebarth.
11. J. Harte, "Water resource constraints on energy development" in California Water Planning and Policy, E. Englebert, ed., pp. 65-87, University of California, Water Resources Center, Davis, CA (1979).
12. J. Harte, "Water constraints on energy development: A framework for analysis," presented at the IIASA-RSI Conference on Systems Aspects of Energy and Mineral Resources, Laxenburg, Austria, July 1979.
13. K. T. Perez, G. M. Morrison, N. F. Lockie, C. A. Oviatt, S. W. Nixon, B. A. Buckley, and J. F. Heltsche, "The importance of physical and biotic sealing to the experimental simulation of a coastal marine ecosystem," Helgolander wiss, Meereunters, 30, 144-162 (1977).
14. M. E. Pilson, G. A. Vargo, and C. A. Oviatt, "Nutrient cycles in MERL microcosms," Proceedings from the symposium on microcosms in ecological research, John Giesy, ed., to be published (1979).
15. D. Schneider, a graduate student in the Energy and Resources Program, will carry out this work.
16. P. Gleick, a graduate student in the Energy and Resources Program, will carry out this study.

TREATMENT METHODS FOR REMOVAL OF BORON FROM BRINES*

W. Garrison and S. L. Phillips

INTRODUCTION

The presence of boron in geothermal brines at concentrations ranging up to several hundred ppm[1] represents a potential hazard to plants and crops. Boron in the form of boric acid and its water soluble salts is an essential trace element in the normal growth of all higher green plants.[2] However, as is the case with most trace-element nutrients, excessive amounts become toxic and lead to plant death. Some boron-sensitive plants and crops show toxic effects when irrigated with water containing boron concentrations as low as 1 ppm.[3] The project has as its objective a study of the mobility and sorption of boron through soils. The goals are: (1) to analyze soil samples for their boron content, (2) to study the sorption of boron by soils from waters such as geothermal brines, and (3) to assess treatment methods for boron removal.

ACCOMPLISHMENTS DURING 1979

A survey was completed of the available data for treatment methods to remove boron from geothermal brines.

The development of economically feasible methods for removal of boron at the ppm level from waste waters--industrial, agricultural and geothermal--is still in progress.[1,3,4,5] Various approaches have been employed with varying degrees of success as outlined below.

Adsorption of boric acid onto hydrated aluminum oxide and iron oxide sites in clays and clay-components of soils could provide an effective method for boron removal. Unfortunately, the adsorptive capacities of clays and related materials are low (0.1 mg/gm) even under optimum conditions. From the plant engineering standpoint, prohibitively large amounts of adsorbate would be required in most cases. However, adsorption processes are an important consideration in the removal of boron by soils in situ following a spill or other accidental contamination.

Precipitation methods commonly used in the water treatment industry, e.g., lime-magnesium, require 13 mol mg/mol B and are costly because of the large quantities of chemicals required. In addition, the conventional sedimentation and biological treatment processes employed in sewage treatment plants have little or no effect on boron levels.

Reverse osmosis (through cellulose acetate) would appear to have limited application in the removal of boric acid from waste waters. At an operating pressure of 500 psi, only 20-30 percent of the boron in sea water is rejected as compared to over 95 percent of the sodium chloride.

Of the waste water treatment processes, those involving ion-exchange resins appear to be the most promising. It is recognized, of course, that the complete de-ionization of water by strongly basic ion-exchange resins such as Amberlite IRA-400 and Dowex 2, does not represent an economical approach for removal of boron from waters containing appreciable concentrations of other exchangeable anions such as chloride ion. However, the strongly basic resins might be employed economically in the removal of boron from geothermal steam condensates which are relatively low in total salinity. Similarly, the Amberlite IRA-400 and Dowex 2 resins could be used in the removal of boron from brackish waters previously treated by one or more reverse-osmosis cycles.

However, it appears that the most direct and economical method for removal of boron from geothermal brines may be the use of the recently developed boron-specific resin, Amberlite XE-243. This is a crosslinked microreticular gel-type polymer derived from the amination of chloro methylated styrene-divinyl benzene with N-methylglucamine. The resin exhibits equilibrium boron capacities in the range 5 mgB/gm with good hydrodynamic properties and chemical stability. The boron selectivity of Amberlite XE-243 is not affected by high concentrations of various other salts including sodium chloride. It has been estimated that total costs for the lowering of boron concentrations from 10 ppm to 1 ppm in irrigation waters would be below $0.03 per thousand gallons. One estimate for removal of both boron and arsenic from geothermal brines using XE-243 is $2,884,000 for 1976 dollar values.[5] Alternative methods such as adsorption onto activated carbon, bauxite and alumina should be studied.[1]

FOOTNOTE AND REFERENCES

*This work was supported by the Lawrence Berkeley Laboratory Director's Funds.

1. W.-W. Choi and K. Y. Chen, "Evaluation of boron removal by adsorption on solids," Env. Sci. Technol., 13 (2), p. 189-196 (1979).
2. J. E. Bonner and J. E. Varner, Plant Biochemistry, Academic Press, New York, NY (1976).
3. F. T. Bingham, "Boron in cultivated soils and irrigation waters," Adv. Chem. Ser., 123, p. 130 (1973).
4. Chemical Technology and Economics in Environmental Perspectives, Task II - Removal of Boron from Wastewater, EPA-560/1-76-007, Environmental Protection Agency, Office of Toxic Substances, Washington, D.C. 20460, April 1976.

5. D. C. Christiansen and J. A. McNeese, "Removal of arsenic and boron from geothermal brines," *Proceedings of 32nd Annual Purdue Industrial Waste Conference*, May 10-12, 1977 (Battelle Northwest Laboratories, Richland, WA 1977).

ORGANIC COMPOUNDS IN COAL SLURRY PIPELINE WATERS*

A. S. Newton, J. P. Fox, and R. Raval

INTRODUCTION

In order to meet the expected increase in use of coal, especially low-sulfur western coals, a large increase in transport capacity will be necessary. Much of this increased capacity will be met by the construction of coal slurry pipelines. At present only one coal slurry pipeline is in operation but several others are expected to be constructed soon.

The transport of coal by slurry pipeline involves the grinding of coal with water in a rodmill to a fineness such that the resulting slurry remains suspended during pipeline transport. A slurry of about 50 percent can be transported by pipeline. The coal user recovers the coal by centrifugation and/or filtration and the recovered moist coal can be burned directly in a power plant. The recovered water must be disposed of in some manner if it is not used as make-up in the power plant. In the Black Mesa pipeline system and Mohave power plant, the moist coal from the centrifuge is burned directly. The water from the centrifuges is further filtered and used as make-up cooling water. Washings from the filters are evaporated in watertight ponds, and no water is released to the aquatic environment.

Because the disposal of pipeline water in a closed system may not be possible in all situations, it is important to have information on what contaminants might be present in the recovered water if proper treatment is to be made before disposal into the aquatic environment. A bibliography of coal-water interactions has been compiled. Several studies of coal-water interactions have been made, either in glass laboratory shaker bottles,[1,2] or in a closed pipeline test loop.[3] These studies have been principally concerned with the leaching into the water of inorganic species such as compounds of the elements As, Pb, Cr, Cd, Mn, Zn and Hg, and the ions nitrate, sulfate and carbonate. Little work has involved the study of the organics in the water. Godwin and Manaham[3] found that leaching of lignites and sub-bituminous coals release humic acids into water and were concerned about possible complexing of metals by the humic acids. Humic acids were estimated to be equal to the chemical oxygen demand (COD) of water. Other studies on the drainage water from coal mines, water from washing coal, and drainage from open coal piles are related but are not exactly equivalent since in these cases the overall system is oxidizing because of the presence of air. In wet grinding coal in a steel rodmill, hydrogen gas is produced and the system rapidly becomes reducing. Possible interactions of nascent hydrogen can result in the fragmentation of organic groups from the surface of the coal to produce organic compounds which may remain in the water. Because of the structure of coal, polynuclear aromatic hydrocarbons and phenolic compounds are expected of this interaction.

Expected concentrations of organic compounds in slurry water is limited to the respective solubility of each compound in water. For phenolic compounds this may be quite high, but for hydrocarbons such as phenanthrene, pyrene, etc., the solubility in water has been shown to vary from 2,700 ppb to less than 1 ppb (micrograms/liter) as shown in Table 1.[4] It is possible that the concentrations can exceed these solubility values if organic compounds are absorbed onto colloidal material or very fine particulates in the water. It is also possible that the coal itself can act as an absorbent for these organic compounds and that the concentrations in pipeline water may be well below the solubilities.

Table 1. Solubilities of some polynuclear aromatic hydrocarbons in water.

Compound	Solubility in micrograms/liter
(anthracene)	89 (@ 20°)
(phenanthrene)	2700 (@ 20°)
anthracene	75
phenanthrene	1600 (1650)
naphthalene	1.0
chrysene	1.5
5-Me chrysene	62
6-Me chrysene	25
1,2 - Benzanthracene	11
Triphenylene	38
Pyrene	165
Perylene	0.5
Picene	2.5
3,4 Benzpyrene	9.0 (4.0)

EXPERIMENTAL

An industrial rodmill for the preparation of coal slurries is some 4 meters in diameter and 5.5 meters long. The average residence time of coal in a continuous mill is only a few minutes. A laboratory batch mill cannot exactly duplicate all of these plant conditions, but a laboratory mill which does duplicate some of them is shown in Figs. 1 and 2. It is made from a 9 inch long piece of 8 inch O.D. seamless steel pipe. Inside are welded 1/2 x 1/2 inch steel bars to carry the rods and provide a good grinding action. Five one inch diameter steel rods are adequate. The cover is sealed with an "O" ring, and it also contains a flush valve through which gas samples can be removed from the rodmill. The net volume of the mill for coal, water and gas space is 5278 ml. The mill is driven on a jar mill drive, Fisher Scientific Co., Model 753 RM. The drive was modified to reduce the speed by adding an additional 4:1 reduction pulley system. With the modified drive the rodmill is rotated at 28 RPM.

The coals to be investigated are Black Mesa coal and Illinois No. 6 coal obtained from the Peabody Coal Company, and Wyodak Roland Seam Coal obtained from Wyodak Resources.

The procedure in the first preliminary experiments has been to add the coal to water and contact with agitation for 16 - 18 hours. The slurry was centrifuged in glass bottles. The water was further filtered. The filtered water was extracted with nano-grade hexane and the hexane extract concentrated to a 1 - 2 ml volume in a vacuum evaporator. The sample volume was further reduced to a few microliters by evaporation with a stream of nitrogen gas.

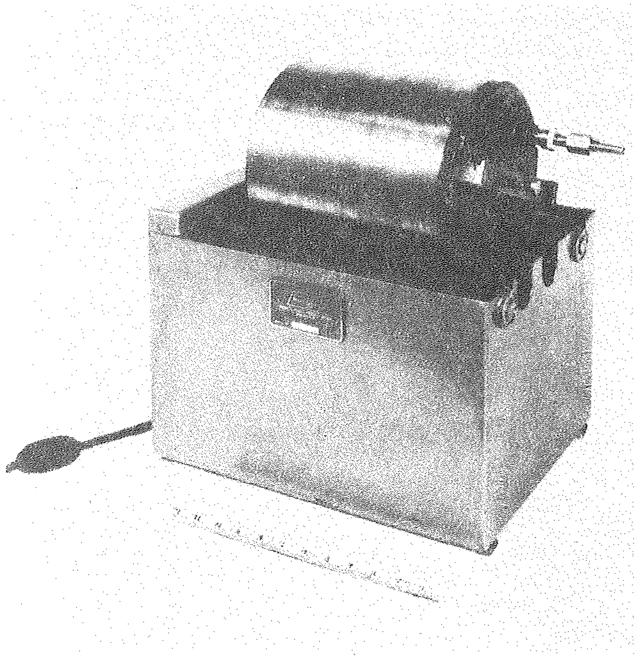

Fig. 1. Photograph of laboratory rodmill and its drive for the preparation of coal slurries. (XBB 790-15385)

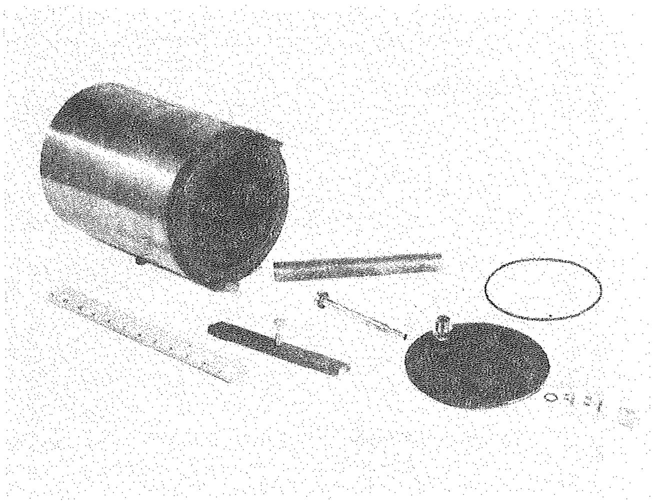

Fig. 2. Photograph of laboratory rodmill in exploded view showing positioning of rods, cover and valving system. (XBB 790-15386)

The concentrated sample was studied using a Finnigan Model 4023 GC/MS. A 30 meter OV-101 capillary GC column was used with Grob injection. This system and method gives excellent identification of many components, but only an approximate quantitative result can be cited. At the present stage of the investigation, the citation of only approximate concentrations is justified.

RESULTS AND DISCUSSION

Three preliminary experiments, the results of which are shown in Table 2, have been performed. Samples Coal A and Coal C were made up with Wyodak coal and laboratory de-ionized water. The results show gross contamination with phthalate esters and squalene. As shown in Table 3, the phthalate esters are traceable to the de-ionized water which is in contact with polyvinyl chloride pipes. The squalene probably arises from fingerprints. These results illustrate the extreme care that must be taken in obtaining and analyzing samples.

The sample Rodmill-1 (Table 2) shows much less contamination than did the earlier samples made with de-ionized water. The low concentrations of hydrocarbons observed in sample Rodmill-1 may be residual contamination from traces of oil which were not removed in cleaning the rodmill but which should gradually be eliminated with use of the rodmill. Water must, however, be distilled from alkaline permanganate to eliminate all organic compounds from that source. No impurities were observed in the hexane.

In Table 4 the gases present in sample Rodmill-1 are shown. Hydrogen, some CO_2, and the oxygen depleted air initially present in the sample were found. In this sample some gas was lost because of excessive pressure build-up. No methane was detected.

Table 2. Organic compounds found in coal slurry water.

Compound	Concentration in p.p.b. in water		
	Coal A[1]	Coal C[2]	Rodmill-1[3]
Diethyl Phthalate	5.0	0.15	⩽0.005
Dibutyl Phthalate	15	2.8	0.15
Hexanedioic acid ester unidentified (not a phthalate)	--	--	0.15
Di-iso-Octyl Phthalate	3.0	0.25	0.05
Squalene	0.3	0.007	0.10
Hydrocarbons C_{15}-C_{30} (each)	⩽0.1	<0.015	<0.005

[1] Coal A sample was 250 gms Wyodak Coal (<100 mesh) shaken with laboratory de-ionized water in a 500 Ml glass bottle. Hexane extract of water analyzed.

[2] Coal C sample was like Coal A except 500 gms lump coal used with 500 Ml water and 100 gms 1 cm steel ball bearings added. Hexane extract of water analyzed.

[3] Rodmill-1 sample was 2300 gms of Wyodak coal ground with 2500 cc LBL tap water (EBMUD water) for 16 hours. Hexane extract of water analyzed.

Table 3. Blank determinations of water and hexane used.

	P.P.B. Impurities In:			
	Deionized water			
Compound	A	B	Tap water	Hexane
Diethyl phthalate	--	--	--	--
Dibutyl phthalate	0.1	0.6	0.001	<0.001*
Hexanedioic acid ester	--	1.6	--	--
Di-iso-Octyl phthalate	14	12	0.004	<0.001*
Squalene	0.3	10	--	<0.001*
Hydrocarbons	--	--	--	--

*No contamination observed at this sensitivity level.

Table 4. Analysis of gases from sample rodmill - 1.

Compound	% Compound	Amount of Gas, Millimoles
H_2	78.9	97
N_2	19.3	24
O_2	0.0	(-6)[a]
Ar	0.24	0.3
CO_2	1.51	1.8

[a] This amount of O_2 was depleted from the air initially present.

The results presented here show that water from a slurry with Wyodak sub-bituminous coal contains no hexane-extractable contaminants attributable to the coal in amounts larger than about 0.1 ppb. In the present experiments phenols and other polar compounds were not determined and volatile compounds such as benzene and toluene would have been lost in the hexane concentration step.

FOOTNOTE AND REFERENCES

*This work was supported by the Fossil Fuel section of the Department of Energy.

1. J. W. Moore, "Water quality aspects of coal transportation by slurry pipeline," Proc. 4th Int. Tech. Conf. on Slurry Transportation, Las Vegas, Nevada, March 28-30, 1979.

2. O. L. Anderson, M. B. Rogozen, W. L. Margler, P. Mankiewicz, and M. H. Axelrod, "Water pollution control for coal slurry pipelines," Institute of Geophysics, UCLA and Science Applications, Inc. Report SA 1-068-79-516, June 30, 1978.

3. J. Godwin and S. E. Manahan, "Interchange of metals and organic matter between water and sub-bituminous coal or lignite under simulated coal slurry pipeline conditions," Environmental Science and Technology, 13, 1100 (1979).

4. H. Seidell and W. F. Linke, "Solubilities of Inorganic and Organic Compounds," p. 758, supp. to 3rd Ed. D. Van Nostrand Co., NY (1952).

PROTECTION OF GEOTHERMAL BRINE PIPING USING ELECTRODEPOSITED ORGANIC LININGS*

S. L. Phillips, R. G. Clem, and S. G. Chang

INTRODUCTION

It is basic to progress in utilization of geothermal energy that adequate and economic materials be available for commercial use. This is true in all aspects of the problem: piping to transport the corrosive hot brine and steam; heat exchangers to transfer heat for driving turbines and for heating structures; drilling of exploratory injection and production wells; measuring flow rates, temperatures and pressures; and establishing material specifications and standards. The present state of the art is far from satisfactory, however, with unprotected or expensive metals being widely used for handling hot brine and steam. Corrosion and scale encrustations are encountered in all geothermal plants; their presence adversely affects plant lifetimes and power output. The problem is sufficiently serious that the use of expensive materials such as type 316 stainless steel and titanium base alloys may be required to insure long term operation of power plants and direct utilization facilities. However, a real economic gain will be realized if the less expensive materials such as carbon steel, which has been used as the primary material of construction in piping and heat exchangers, can be used to handle hot brine and other aerated geothermal fluid.[3,4,5]

A commonly used technique to control corrosion and to increase the resistance of carbon steel piping to erosion, corrosion, and scaling is to use protective coating. These coatings are applied to the metals in a number of ways; the films may be organic or metallic. A more recent approach to this problem, which has found acceptance by industrial manufacturers for a variety of applications, is the electrodeposition of organic films.

The electrodeposition of organic coatings from aqueous systems is widely used, for example, to paint automobiles for corrosion protection. The advantages of this method include ease of automation; uniformity of film thickness; penetration of the film into cracks, flaws and other not easily accessible places; high utilization of both resin and pigment (greater than 95 percent); and the short time needed between film deposition and rinsing. Besides these, it is possible to electrodeposit resins such as fluorocarbons (e.g., Teflon) which are included among those coatings with superior resistance to chemicals, abrasion, high temperature, and friction.[1,2,6,7]

Because of these considerations, work was initiated in September, 1979, to evaluate electrodeposition procedures for coating and lining the inside of piping with Teflon for geothermal energy applications.[8]

ACCOMPLISHMENTS DURING 1979

During the one month of funding, laboratory work was centered mainly on assembling equipment, obtaining commercially available resin dispersions, and proving the concept of lining the interior of metal piping with an electrodeposited organic film. For reasons stated, Teflon was selected as the initial resin for the pipe lining. It is impossible to deposit Teflon onto metals such as copper with satisfactory adhesion; Hycar (acrylonitrile/butadiene) is co-deposited thereby providing a marked improvement in adhesion.[1]

Experimental

The electrodeposition was carried out under conditions of constant applied voltage using a Power Designs Inc. Model 2050 power supply. For coating the interior of copper pipes, collars were machined from bulk Teflon to fit over each end and to contain the aqueous coating bath. The copper piping was 2.5 cm (1 inch) diameter, 15 cm (6 inch) long, hard drawn seamless, ESR NC No. 234. A 1.3 cm (0.5 inch) diameter copper rod was inserted through the center of the Teflon collars to serve as the cathode during electrodeposition. Copper coupons for deposition studies were cut from 0.025 cm thick OFHC cold rolled, half-hard copper sheeting, CDA alloy 110, ESR 120; typical dimensions of the coupons were 2 cm x 2 cm. A 0.5 cm x 2 cm long tab was left on each coupon to provide electrical contact via an alligator clip with a moving anode bar. The anode bar was moved back and forth about 2.5 cm (1 inch) at a rate of six cycles per minute using a synchronous motor. This moving bar provided agitation and reduced concentration polarization effects traceable to the dispersed material during deposition. A 1000 ml stainless steel beaker served simultaneously to contain the coating bath and as the cathode for deposition onto the coupons.

Both dispersions were proprietary and were used as received. The polytetrafluoroethylene was E. I. DuPont Teflon 30; the acrylonitrile/butadiene copolymer was B. F. Goodrich Hycar 1561. Table 1 lists selected properties of the dispersions; Table 2 consists of selected properties of bulk Teflon. Electrocoating baths were prepared by mixing known weights or volumes of each dispersion.

Table 2. Selected properties of bulk Teflon.

Resin	Thermal conductivity, W/m.K	Rockwell Hardness	Specific gravity
Teflon	2510	D50 to D60	2.1 to 2.3

Electrocoated Coupons

Copper coupons were cleaned by scrubbing with a proprietary powder cleaner, followed by washing, etching for 10-15 minutes in ammonium persulfate solution (160 g/l), and rinsing with distilled water. The presence of a continuous water film on the resulting salmon-colored copper was taken as the criterion for a cleaned surface. After electrocoating, the coupons were rinsed with distilled water, dried 1-2 hours at ambient temperature, heated to 75-80°C for 4-5 hours; then placed in a muffle furnace at 300°C for 15-20 minutes. (Blisters developed on coupons which were placed in the muffle furnace directly after coating.)

The Teflon/Hycar film was bluish-green in color after deposition; the color was unchanged on heating to 150°C. However, heating to 300°C resulted in a black, smooth film. Bending a coated coupon 180° caused white cracks to appear in the coating, but the film was not readily dislodged from the copper by picking with a sharp knife. This indicated good adhesion. Table 3 shows the variation in film thickness, ℓ, with time, t, for a series of coated coupons. The constancy of the ratio $\ell/t^{1/2}$ indicates diffusion-controlled film buildup.

Electrocoated Piping

The inside of the copper pipe was cleaned by scrubbing with a powder cleaner using a bristle brush, followed by water rinsing. The rinsed tube was then filled with about 55 ml of ammonium persulfate solution and allowed to sit for 10-15 minutes. After this time, the persulfate was decanted, the pipe rinsed with distilled water, and the Teflon/Hycar resin dispersion added. After 2-3 minutes, the two leads to the power supply were connected and 10 volts applied for 2 minutes. Gas evolution was evidenced by foaming at the top

Table 1. Selected properties of water-based Teflon 30 and Hycar 1561 dispersions from manufacturer's literature.

Resin	Solids, %w	Particle size, micron	pH	Wetting agent, %w	Viscosity, centipose	Specific gravity
Teflon 30	60	0.05 to 0.5	10	8	20	1.50
Hycar 1561	41	--	10	-	27	1.00

Table 3. Effect of deposition time on coating thickness using moving anode bar and copper coupons. Deposition voltage was 40 volts; dispersion was composed of 600 ml of Teflon 30 mixed with 325 ml of Hycar 1561.

Coating time sec. (t)	$t^{1/2}$	*Coating thickness, cm. (1)	$1/t^{1/2}$
10	3.16	0.010	0.0032
30	5.48	0.0178	0.0032
60	7.75	0.0229	0.0030
120	10.9	0.0305	0.0028
180	13.4	0.0381	0.0028
300	17.3	0.0457	0.0026

*Coating thickness measured with a micrometer; copper coupon was 0.025 cm thick.

of the pipe. Following deposition, the aqueous dispersion was decanted, the tube thoroughly washed with distilled water, and allowed to dry at ambient temperature for several hours. The tube was then heated to 300°C for 15-20 minutes, after which time a smooth and adherent black lining was formed.

The plating current was monitored during deposition; see Table 4.

Table 4. Variation in current with time for electrolined copper piping. Applied potential, 10 volts.

Time, sec.	5	30	60	90	120
Current, amp.	1.8	1.4	0.8	0.5	0.4

The results of this work show that copper piping can be lined with an organic film comprised of Teflon and Hycar using electrodeposition procedures.

PLANNED ACTIVITIES FOR 1980

Proposals will be submitted to request funding for a project centered on developing both materials and processes for lining steel piping with organic resins using electrodeposition methods.[9] The result of this proposed work will consist not only of lined steel piping, but also of test data on the corrosion, erosion and scaling protection afforded to the piping in contact with geothermal hot brines and vapor systems. Methods for joining shorter lengths of piping and for coating angular bends (e.g., elbow joints) will form part of the proposed work. Both materials and deposition processes will be characterized using modern instrumental methods.

FOOTNOTE AND REFERENCES

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1. S. L. Phillips and E. P. Damm, "Electrophoretic deposition of resins at soluble metal anodes," J. Electrochem. Soc., 118(2), 1916-1919 (1971).
2. F. Beck, "Fundamental aspects of electrodeposition of paint," in Progress in Organic Coatings, 4, 1-60 (1976).
3. Alternate Materials of Construction for Geothermal Applications, BNL 50979, October - December 1978, Progress Report No. 17, Brookhaven National Laboratory, Upton, NY 11973.
4. American Society for Testing and Materials Task group on Geothermal Seals, Proceedings of Technical Seminar, CONF-7806141, April 1979, Boston, MA, June 27, 1978.
5. Materials Selection Guidelines for Geothermal Power Systems, ALO-3904-1, D. W. DeBerry, P. F. Ellis and C. C. Thomas, Radian Corp., Austin, TX 78766, September 1978.
6. E. B. Ewan, "Coatings that can really take it," Materials Engineering 89, 38-42 (1979).
7. A. G. Roberts, Organic Coatings, Building Science Series 7, National Bureau of Standards, Washington, DC (1968).
8. D. R. Sherwood and D. J. Whistance, The Piping Guide, Part 1 (1973).
9. Proposal submitted to DOE Division of Geothermal Energy, January 30, 1980.